



## Multiple-locus variable-number tandem-repeat analysis (MLVA) typing of *Salmonella* Typhimurium DT41

Litrup, Eva; Christensen, Henrik; Nordentoft, Steen; Davies, R.H.; Helmuth, R.; Møller, Eva; Bisgaard, Magne

*Publication date:*  
2010

[Link back to DTU Orbit](#)

### *Citation (APA):*

Litrup, E., Christensen, H., Nordentoft, S., Davies, R. H., Helmuth, R., Møller, E., & Bisgaard, M. (2010). *Multiple-locus variable-number tandem-repeat analysis (MLVA) typing of Salmonella Typhimurium DT41*. Poster session presented at International Symposium Salmonella and Salmonellosis 2013, Saint-Malo, France.

---

### General rights

Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

- Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
- You may not further distribute the material or use it for any profit-making activity or commercial gain
- You may freely distribute the URL identifying the publication in the public portal

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

# Multiple-locus variable-number tandem-repeat analysis (MLVA) typing of *Salmonella* Typhimurium DT41

<sup>1</sup>Eva Litrup, <sup>2</sup>Henrik Christensen, <sup>3</sup>Steen Nordentoft, <sup>4</sup>R.H.Davies, <sup>5</sup>R. Helmuth, <sup>1</sup>Eva Møller Nielsen, and <sup>2</sup>Magne Bisgaard

<sup>1</sup>Statens Serum Institut, 5 Artillerivej, Copenhagen, <sup>2</sup>Department of Veterinary Disease Biology, University of Copenhagen, Frederiksberg, Denmark, <sup>3</sup>National Veterinary Institute, Technical University of Denmark, Aarhus, Denmark, <sup>4</sup>Veterinary Laboratories Agency Weybridge, UK, <sup>5</sup>Federal Institute for Risk Assessment, Berlin, Germany

## Introduction

The Danish Salmonella Control Programme aims at eradication, and does not allow vaccination at any level. As a result, the occurrence of *Salmonella*-positive broiler flocks at processing has decreased significantly during the past decades. Broilers found to be infected before slaughter are slaughtered at a single slaughterhouse under special hygienic precautions and the meat is heat treated (1). One specific phagetype, DT41, of *S. Typhimurium* has been predominant in broiler breeder flocks and has been found in 13 broiler parent flocks during the last ten years (unpublished results). The reason for such a high frequency remains to be identified.

## Material and methods

33 isolates from Danish poultry production collected from 12 outbreaks during a 12 year period and 38 unrelated isolates of veterinary origin from Germany and England, as well as 30 human isolates from Danish patients during 1998-2008 were included for comparison. Phage typing was performed according to the phage typing scheme developed by Callow (2) and extended by Anderson (3). MLVA was performed as described previously (4). MLVA repeats based on 5 alleles were calculated according to the method described previously (5), and a neighbour joining dendrogram was calculated in ClustalX (6) based on a matrix obtained by coding each MLVA allele as a discrete character and taking the mean of all five alleles. Bootstrap analysis was performed from ClustalX.

## Results and discussion

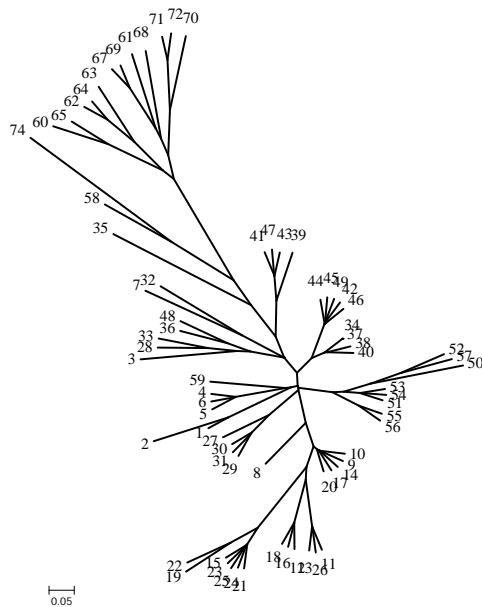
Cluster analysis of the MLVA similarities showed one large cluster and at least 14 minor clusters, plus 26 more unrelated isolates (Fig. 1). The major cluster consisted of 20 of the 30 human isolates and isolates from pigs, pig meat and a reptile. The other clusters and singletons included the remaining animal isolates, including all the Danish outbreak isolates.

Amongst Danish chicken isolates the same MLVA types were never observed in different years (Table 1), suggesting that DT41 clones did not persist in the Danish production system but had an outside source. Transmission from breeder rearing farms to egg producing adult parent stock was not demonstrated. A broiler flock originating from one breeder flock demonstrated the same MLVA type (9) as the parents (Table 1). However, no other Danish broiler farm has tested positive for DT41 during the past decade (1). In only a single case was the same MLVA type (38) demonstrated among both human isolates and isolates from broiler breeders. However, these isolates differed significantly as to time of isolation underlining that poultry does not represent a significant reservoir of DT41 for human beings in Denmark. The lack of observation of the same MLVA type for more than three years (only for type 38 five years) can either be related to instability of MLVA types within the same strain (reversible or irreversible) or to lack of reoccurrence of strains if they have stable MLVA types.

# Conclusion

Transmission from Danish broilers to humans was not demonstrated, neither was the transmission from rearing farms to broiler breeder farms. Sources of infection at broiler breeder farm level remained unidentified. The wild fauna is considered to represent a risk factor for poultry, since DT41 is often identified in wild birds, however, this still needs to be substantiated by further studies.

Fig. 1. Neighbour joining cluster analysis based on a similarity matrix of diversity between the MLVA profiles (numbered).



# References

1. Anon (2009) Annual report on zoonoses in Denmark 2008. National Food Institute, Technical University of Denmark ([www.food.dtu.dk](http://www.food.dtu.dk)).
2. Callow, B.R. (1959) A new phage-typing scheme for *Salmonella typhi-murium*. *J Hyg (Lond)*. 57, 346-359.
3. Anderson, E.S., Ward, L.R., de Saxe, M.J., and de Sa, J.D.H. (1977) Bacteriophage-typing designations of *Salmonella typhimurium*. *J Hyg (Lond)*. 78, 297-300.

Table 1. Danish chicken isolates of *Salmonella Typhimurium* DT41 from broiler breeder farms investigated by MLVA.

MLVA Type	Year	Broiler breeder farm number
11	1998	1
13	1998	1
13	1998	1
13	1998	1
5	1999	3
9	1999	3
9	1999	3
9	1999	broiler farm from 3
6	1999	2
45	2000	5
53	2002	6
14	2002	7
12	2003	8
12	2003	8
26	2004	8†
31	2004	9
38	2006	10
40	2006	11
28	2006	11
38	2006	9
39	2006	10
49	2007	13
51	2007	12
43	2008	14
36	2009	1†
36	2009	1†
36	2009	1†
36	2009	1†
36	2009	1†
36	2009	1†

† The 2nd flock on the same farm found positive.

4. [Torpdahl, M.](#), [Sørensen, G.](#), [Lindstedt, B.A.](#) and [Nielsen, E.M.](#) (2007) Tandem repeat analysis for surveillance of human *Salmonella* Typhimurium infections. *Emerg Infect Dis*. 13, 388-95.
5. Larsson, J., Torpdahl, M., Petersen, R.F., Sørensen, G., Lindstedt, B.A. and Nielsen, E.M. (2009) Development of a new nomenclature for *Salmonella* Typhimurium multi-locus tandem repeats analysis (MLVA). *EuroSurveillance* 14, (15)
6. Thompson, J.D., Gibson, T. J., Plewniak, F., Jeanmougin, F. and Higgins, D. G. (1997) The CLUSTAL\_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucl Acids Res* 25, 4876-4882.